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ARTICLE

# Polymer Chemistry at the Living-Material Interface

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Engineered living materials (ELMs) integrate living functionalities into synthetic materials. Although most ELM studies emphasize cellular functionality, the polymer matrix represents an equally powerful design space: not merely a passive scaffold but a primary tool for programming living material behavior. Synthetic polymers can be designed to tune bulk mechanical properties, mediate dynamic responses, enforce biocontainment, and bridge biological and synthetic domains through engineered polymer-cell interfaces. This Perspective establishes key design principles that emerge when polymer chemistry and living cells are treated as an integrated system, emphasizing how molecular control over the living-material interface governs the mechanical, functional, and dynamic properties of next-generation biohybrid and biocomposite materials.





## Introduction

Natural living materials possess a remarkable range of mechanical and functional properties, refined through evolutionary pressures over millions of years. They combine renewable composition, degradability, and adaptable behaviors such as self-healing and multimodal responsiveness, properties that remain difficult to replicate in synthetic systems. This disparity has motivated the development of engineered living materials (ELMs), which integrate living cells into material architectures to introduce biological function.[1-3] In these systems, embedded cells enable self-repair, environmental sensing, and catalysis, offering capabilities often inaccessible to abiotic materials. Beyond these biologically derived capabilities, ELMs also present a materials landscape at the interface of living and synthetic matter, where cell selection, macromolecular scaffold design, and the polymer-cell interface collectively govern emergent material mechanics and function.

Fabrication of ELMs generally follows two primary routes.[3] In bottom-up approaches,[4] cells are engineered to produce and assemble structural components such as protein nanofibers,[5-9] extracellular polysaccharides,[10, 11] and mineralized biofilms.[12] In contrast, top-down strategies embed cells within polymeric networks such as hydrogels.[13, 14] In practice, the choice of route is largely dictated by cell compatibility and the requirement to maintain viability within artificial environments. Expanding the design space of ELMs will therefore require identifying polymer systems that are intrinsically cell-compatible, or alternatively, selecting cell types tolerant of synthetic material environments.

Collectively, ELM design can be understood across three coupled levels: (i) the polymer scaffold that governs bulk mechanics and processing; (ii) the polymer-cell interface that encodes molecular interactions and emergent behavior; and (iii) the biological chassis selection that determines the intrinsic functional capabilities and environmental tolerance of the living component (e.g., vegetative cells versus spores).

In this Perspective, we argue that synthetic polymer chemistry provides a powerful framework for tailoring the mechanical, structural, and functional properties of ELMs (Figure 1). Through early studies that establish key design principles for future development, we emphasize substantial and underexplored opportunities for polymer chemists in this space. Advances in polymer synthesis have established an extensive toolbox of design strategies that enable precise molecular control over cell-material integration and emergent properties. Other approaches to ELM construction, such as bottom-up approaches rooted in synthetic biology and application-driven developments, have been summarized elsewhere;[1-4, 15] here, we focus on the design logic that emerges when polymer chemistry and living cells are treated as an integrated system.

We first outline the broader design space of ELMs accessible through cell-compatible polymers and formulation methods that support viability and function (Figures 1a and 1c). We then examine how molecular-level interactions (Figure 1b) at the polymer-cell interface can be leveraged to tune material properties (Figure 1d), including stiffness, viscoelasticity, biocontainment, dynamic phase behavior, recyclability, and stimuli-responsiveness. Next, we highlight the incorporation of polymer-compatible bacterial spores as a robust living component, along with key molecular design parameters that govern their integration (Figure 1c). Lastly, we discuss emerging frontiers at the intersection of synthetic chemistry and synthetic biology, where rapidly expanding toolsets are enabling unprecedented control over the living-material interface.

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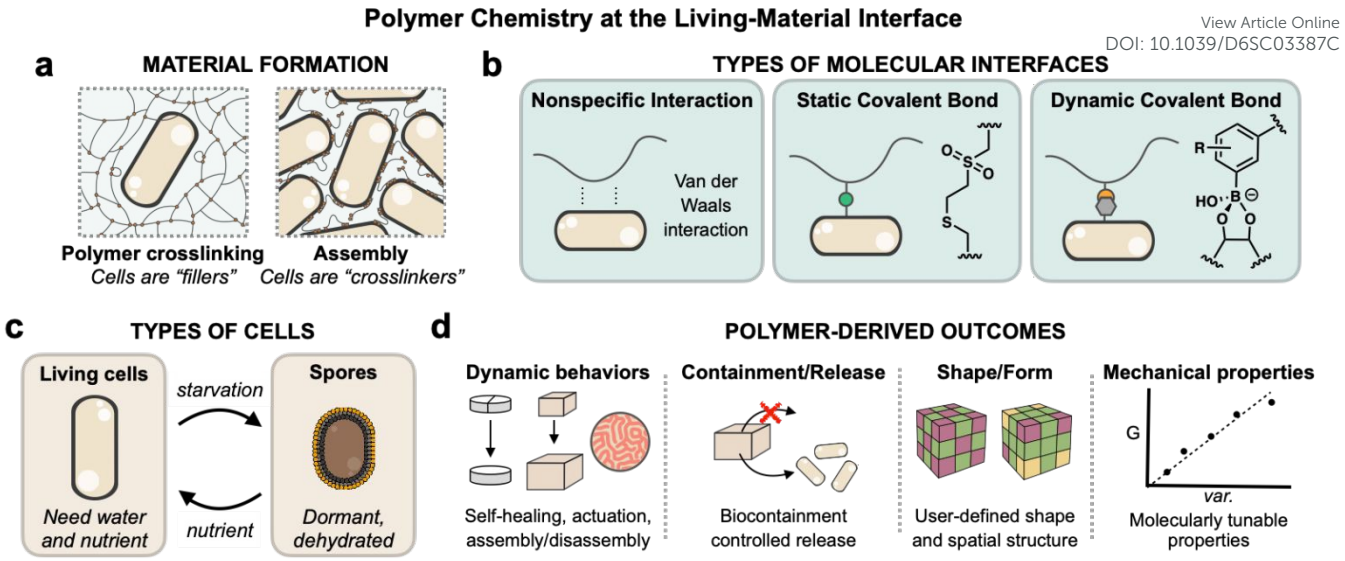


**Figure 1. Overview of this Perspective: Polymer Chemistry at the Living-Material Interface.** (**a**) Two different approaches in material formation with living particles. (**b**) Types of molecular interfaces in polymeric living materials. (**c**) Two different types of cellular particles described in this Perspectives. (**d**) Polymer-derived functions in living materials.

## General design considerations for polymer scaffolds in living materials

A central design consideration for ELMs is supporting cellular viability within the macromolecular scaffold. Consequently, many ELM platforms rely on hydrogel networks and water-soluble polymers that extend conventional cell culture conditions into 3D environments, often without incorporating specific polymer-cell interactions. Viability requirements vary across cell types, including bacterial, fungal, and mammalian systems. However, common constraints for cell survival, such as the availability of water and nutrients and the maintenance of a suitable pH and temperature range, persist across most biological systems.

Cells within ELMs can function either as "fillers" dispersed within macromolecular scaffolds or as active "crosslinkers" that molecularly assemble with synthetic polymers (Figure 1a). This distinction shapes downstream design choices, including polymer architecture, processing conditions, and emergent material behavior. In this Perspective, we discuss both strategies and highlight the unique advantages that come with each construct. Most designs use cells as fillers, processing them into materials under biocompatible conditions. This approach physically decouples cells from the network, prioritizing cell viability and growth in the 3D environment. Accordingly, molecular polarity and crosslink density must be fine-tuned to prevent uncontrolled cell leakage and phase separation. Using cells as crosslinkers overcomes these issues by directly bonding polymers and cells. Polymer-cell crosslinks also introduce bond-specific properties that directly affect the cells' local environment.

Together, these two strategies define a continuum of polymer-cell coupling, ranging from passive physical encapsulation to permanent covalent integration at the living-material interface. Intermediate regimes include supramolecular interactions and dynamic covalent bonds that exchange under mild conditions. Each coupling mode carries distinct consequences for cell mobility, network mechanics, stimuli-responsiveness, and biocontainment. The following subsections illustrate these design principles through examples of cells functioning as fillers, organized by how polymer architecture and fabrication strategy govern material mechanics and function. Subsequent sections contrast these with polymer-cell bonding strategies that afford greater control over living material properties.

### Coupling cellular proliferation to material mechanics

In filler-based designs, where cells are physically dispersed without direct polymer-cell bonds, cell metabolism and growth can become intrinsic mechanical variables that couple biological activity to bulk material behavior. Controlled proliferation, for instance, can induce bulk shape changes whose magnitude depends on scaffold crosslink density. Rivera-Tarazona *et al*. used this mechanism to create living shape-changing materials.[16] Poly(acrylamide) (pAm) hydrogels were prepared by free radical polymerization using an aqueous acrylamide monomer and bisacrylamide crosslinker solution in the presence of *Saccharomyces cerevisiae* yeast cells. Incubation of the living gel in a nutrient medium resulted in cell proliferation-induced volume change of ~201%, significantly higher than controls without encapsulated yeast (~1%). Material stiffness and growth were controlled through crosslink density: Higher crosslink density increased stiffness, but also reduced the volume changes caused by cell proliferation. Kalairaj *et al*. demonstrated that the same proliferation-driven

pressure can be redirected toward material fracture for controlled cell release.[17] The rate of release was tunable by varying the initial cell loading or the crosslink density, and the fracture mechanism could be adapted to release model gram-positive, gram-negative, and fungal probiotics.

**Polymer architecture as an independent mechanical handle**

Crosslink density is one handle to program mechanics, but polymer topology and side chain composition offer independent and complementary means of control. Murphy *et al*. designed shear-thinning 3-arm block copolypeptide star polymers that enabled direct ink writing of materials containing *Escherichia coli*.[18] Poly(benzyl-L-glutamate) blocks supported bacterial viability through hydration and allowed engineered strains to express green fluorescent protein (GFP). Incorporation of poly(L-valine) or poly(L-tyrosine) blocks drove tunable β-sheet formation, modulated stiffness, and conferred hydrogen bond-mediated strain recovery. Covalent crosslinking of methacrylate handles further increased the storage modulus of the poly(L-valine) variant to ~3000 kPa using biocompatible photopolymerization (405 nm). Notably, bacterial growth was more robust in the softer poly(L-tyrosine) construct, providing insight into a potential trade-off between viscoelastic stiffness and cell proliferation.

**Composite fabrication for decoupled mechanical and biological design**

Liu *et al*. reported a wearable *E. coli*-based material constructed by a hybrid hydrogel-elastomer architecture in which polyacrylamide-alginate hydrogels were integrated with polydimethylsiloxane (PDMS) elastomers.[19] Cells and nutrients were loaded into pre-formed microscale cavities within the elastomer and sealed using rapidly curable pre-gel solutions, enabling uniform spatial distribution of cells throughout the composite. In this design, the hydrogel serves a dual role: the covalently crosslinked polyacrylamide network provides stretchability, while the reversibly crosslinked alginate component dissipates mechanical energy, together yielding a tough and highly extensible matrix. The air-permeable PDMS elastomer further maintains cellular viability by supporting gas exchange. *E. coli* encapsulated within the hydrogel-elastomer composite retained >90% viability under humid conditions and in growth media, and engineered strains successfully executed genetically encoded biosensing and production functions, demonstrating that architectural decoupling of mechanical and biological design requirements is a viable strategy for integrating living function into wearable devices.

**Post-fabrication cell incorporation as a biocompatible processing strategy**

The designs above require cells to be present during network formation, exposing them to potentially cytotoxic precursors. For instance, acrylic monomer *N*-isopropylacrylamide (NIPAm) is cytotoxic to the photosynthetic cyanobacterium *Synechococcus elongatus*.[20] Post-fabrication cell diffusion can decouple cell incorporation from network formation entirely, as Tang *et al*. demonstrated using a temperature-responsive Laponite nanoclay-poly(NIPAm) hydrogel. The network undergoes reversible shape deformation across a biocompatible temperature range, driven by poly(NIPAm)'s lower critical solution temperature. The hydrogel was contracted at 37 °C to remove excess NIPAm, then re-swollen at 22 °C in the *S. elongatus* culture and growth medium. The diffusion-based approach resulted in a higher cell density near the surface, with growth occurring throughout the material but concentrated near regions of enhanced light penetration and gas exchange. The resulting pseudo-bilayer generated a local stiffness mismatch, demonstrating that even passive cell loading strategies can introduce structural gradients with functional consequences.

**Bioprinting as a route to spatially programmed architectures**

Bioprinting deposits living and synthetic components in prescribed three-dimensional architectures. By controlling where cells reside relative to chemically and biologically distinct domains, bioprinting can encode anisotropic mechanics, directional response, and compartmentalized function (i.e., division of labor) within a single material. Johnston *et al*. demonstrated this approach by spatially compartmentalizing individual microbial populations within F127-bisurethane methacrylate (F127-BUM) hydrogels to form solid-state bioreactors that produce small molecules and peptides on demand.[21] The temperature-responsive, shear-thinning rheology of F127-BUM enabled the independent encapsulation and extrusion of yeast and bacteria prior to photocuring. Spatial separation of the microbes suppressed the competitive growth dynamics that typically cause one organism to dominate in liquid co-culture, while allowing a cascade reaction in which engineered *E. coli* produced L-DOPA, which was then converted by engineered *S. cerevisiae* into betaxanthins. In this way, bioprinting strategies that integrate cell-compatible chemistries with multi-material fabrication provide a route to spatially program living-material interfaces for improved functionality.

## Molecularly programmable living-material interfaces

Molecular interactions govern the properties of soft materials. Thermoplastics with weak molecular interactions exhibit flexibility, ductility, and processability, while covalently crosslinked thermosets confer stiffness, brittleness, and chemical stability. In gels, polymer-solvent interactions further shape network mechanics. Similar principles also govern biological materials, with cells acting as microscopic fillers within macromolecular scaffolds. Modern synthetic chemistry allows these principles to be extended to ELMs, where interfacial compatibilization between biological and synthetic components can be engineered at the molecular level (Figure 1b). Within this framework, polymer-cell interactions can control cell capture and release, spatial distribution, bulk mechanics, phase behavior, processability, and recyclability. Achieving such control begins with characterizing or modifying the cell surface and tailoring polymer architectures to complement cell-surface chemical features. The following sections develop this

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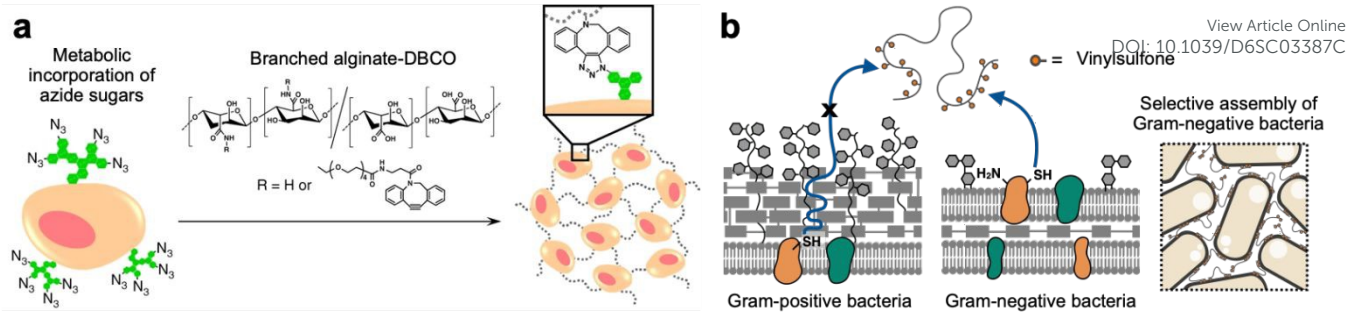


**Figure 2. Examples of Static Covalent Bonding at the Living-Material Interface.** (**a**) Covalently crosslinked polymer-cell network using bioorthogonal click reaction: Metabolically incorporated azide sugars on the cell surface were reacted with DBCO-functionalized branched alginic acid. Adapted from ref. 22, Copyright © 2018, The Authors. (**b**) Selective covalent assembly of vinylsulfone-decorated block copolymers with gram-negative bacteria. Adapted from ref. 23, Copyright © 2026, The Authors.

framework from static to dynamic to dissipative interfaces, tracing how increasing molecular programmability at the polymer-cell boundary unlocks progressively more sophisticated material behaviors.

### Static covalent interfaces

Covalent bonds provide the most mechanically robust and chemically durable interactions available for polymer-cell integration. Crosslinking yields immobilized cellular inclusions within a polymeric matrix and offers a straightforward route to engineering ELMs in which structural rigidity and biocontainment are key objectives. Currently, few such examples exist, likely reflecting the limited tolerance of living cells to irreversible modifications and permanently constrained microenvironments. Nevertheless, investigations of covalent polymer-cell integration offer critical insight into the distinct design constraints that distinguish abiotic polymer networks from living biohybrid materials. Moreover, the design space of ELMs constructed through covalent polymer-cell interfaces may expand with the development of engineered strains capable of maintaining function under artificial or chemically demanding conditions.

Hydrogel ELMs can be constructed by engineering cells to present crosslinking handles on their surfaces. Nagahama *et al*. demonstrated the integration of mammalian cells into a synthetic hydrogel using bioorthogonal click chemistry (Figure 2a).[22] Mouse myoblasts were first metabolically labeled with *N*-azidoacetylmannosamine ($Ac_4ManNAz$) to install azide functionalities on cell surface glycans. Labeled cells were then incubated with dibenzocyclooctyne (DBCO)-functionalized branched alginic acid under physiological conditions (phosphate-buffered saline, 37 °C). Strain-promoted azide-alkyne cycloaddition between the polymer and cell-surface azides yielded percolated polymer-cell networks that formed self-supporting hydrogels, whereas omission of either reactive handle prevented gelation. Rheological measurements confirmed that gel stiffness depended on both the presence and abundance of azide-modified cells, directly correlating bulk mechanical properties with polymer-cell crosslink density. Confocal laser scanning microscopy revealed a uniform cellular distribution, while live/dead staining confirmed cell viabilities exceeding 93%. Importantly, crosslinked cells retained biological function, as evidenced by sustained proliferation and preferential adhesion to collagen. This strategy was further extended to azide-modified lung, femoral muscle, kidney, and heart tissues, all of which formed analogous alginic acid-tissue hybrid hydrogels.

Chemoselective bond formations can be leveraged to facilitate strain-specific ELM assembly based on native cell-surface chemistry. We reported the selective covalent assembly of gram-negative bacteria using vinyl sulfone (VS)-decorated triblock copolymers (Figure 2b).[23] VS reacts selectively with cysteine residues on bacterial surface proteins to form static covalent bonds. While the small molecule VS probe labels both gram-positive and gram-negative bacteria, the triblock VS polymer selectively assembles gram-negative bacteria. We attributed the observed selectivity to differences in bacterial cell-surface structures and macromolecular diffusion barriers. Encapsulated cells retained metabolic function, as demonstrated by melanin production that imparted distinctive optical, mechanical, and biocontainment properties to the composite.

Covalent assembly of ELMs demonstrates that living cells can serve as multifunctional crosslinking nodes within synthetic polymer networks. This molecular assembly approach provides a uniquely well-defined platform in which polymer architecture, crosslink density, and network topology can be systematically varied and directly correlated with bulk mechanical properties. Molecular-level polymer-cell interactions are not passive structural features, but programmable design parameters that govern stiffness, viscoelasticity, and network integrity. These principles find their fullest expression in ELMs constructed using dynamic covalent crosslinks.

### Dynamic covalent interfaces

Self-supporting polymer networks, including many hydrogels and thermosets, rely on covalent crosslinks to confer mechanical robustness and long-term stability. However, static covalent bonds impose fixed network connectivity, limiting adaptability, stress dissipation, and responsiveness – hallmarks of biological materials. Living systems, by contrast, maintain mechanical integrity through dynamic and reversible molecular

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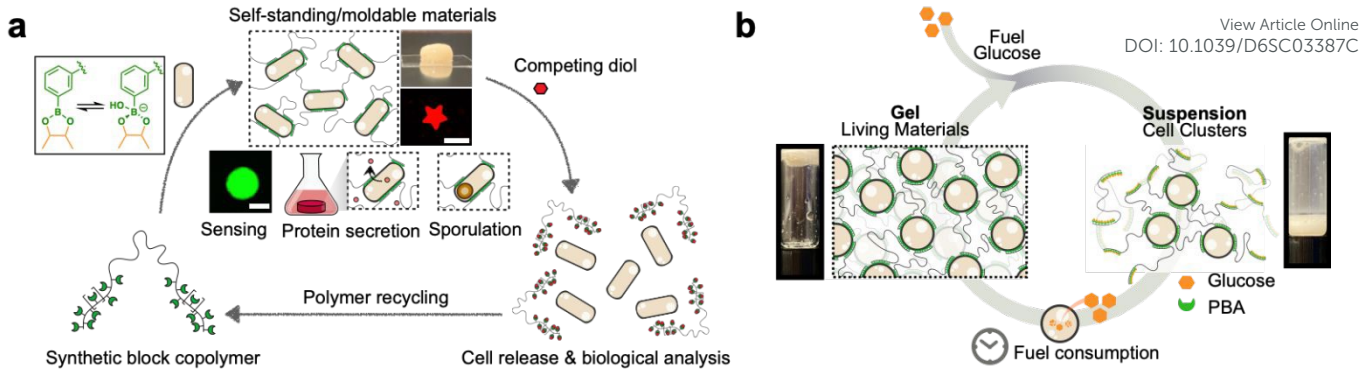


**Figure 3. Examples of Dynamic Covalent Bonding at the Living-Material Interface.** (**a**) Dynamic covalent assembly of boronic acid-functionalized synthetic polymers with a gram-positive bacterium, *Bacillus subtilis*, yielding ELMs with programmable functional properties. Adapted with permission from ref. 28, Copyright © 2022, American Chemical Society. (**b**) Dissipative living materials in which metabolic consumption of glucose fuel drives phase transition. Adapted with permission from ref. 30, Copyright © 2023, American Chemical Society.

interactions that are continuously regulated by cellular activity and extracellular cues.

Dynamic covalent crosslinks can introduce analogous adaptability into synthetic ELMs while retaining molecular precision. These bonds undergo reversible formation, cleavage, and exchange under mild conditions because the relevant activation barriers are accessible at ambient and physiological conditions.[24] At the bulk scale, this translates into time-dependent and stimuli-responsive behavior,[25-27] including stress relaxation, self-healing, reprocessability, controlled cargo release, and viscoelasticity. Importantly, in ELMs, these dynamic processes occur in direct interplay with cellular activity, allowing network connectivity to reorganize in response to cell-mediated changes in the local chemical environment. This coupling transforms cells from passive inclusions into active regulators of material behavior.

We demonstrated a dynamic covalent assembly strategy between boronic acid-functionalized synthetic polymers and a gram-positive bacterium, *Bacillus subtilis*, yielding ELMs with programmable functional properties and recyclable components (Figure 3a).[28] The design rationale centered on cell surface teichoic acids, polyol-rich glycopolymers decorating the cell wall, as endogenous anchoring points for reversible boronate ester bond formation. Small molecule labeling experiments using fluorescent 3-acetamidophenylboronic acid (APBA) probes showed preferential binding to a laboratory strain of *B. subtilis* over a glycosyl transferase knockout mutant of *B. subtilis* or gram-negative bacterium *E. coli*, which lacks teichoic acids, consistent with diol-specific boronate ester formation rather than nonspecific adsorption.

Multivalent polymer-cell interactions and macromolecular diffusion barriers at the interface were probed using APBA-containing polymers synthesized by reversible addition-fragmentation chain-transfer (RAFT) polymerization. Fluorescently labeled APBA polymers coated *B. subtilis* surfaces in proportion to the total number of APBA repeating units, showing that multivalency amplifies dynamic covalent polymer-cell interactions. Moreover, larger polymers promoted the formation of cellular aggregates. Addition of competitive exogenous diols, such as glucose, sorbitol, and fructose, displaced polymer chains from cell surfaces in a competitor-dependent manner observable by fluorescence microscopy. The superior bond exchange efficacy of fructose and sorbitol relative to glucose was consistent with the known thermodynamic preference of boronic acids for diols that adopt a 1,2-cis coplanar conformation, which is particularly prevalent in the furanose form of fructose and in the extended polyol chain of sorbitol.[29]

Viscous aqueous solutions of poly(APBA)-PEG-poly(APBA) telechelic block copolymers underwent gelation upon addition of *B. subtilis* cells, forming self-standing hydrogels in which cells served as dynamic crosslinking nodes. Rheological characterization confirmed a cell-dependent increase in both storage and loss moduli, establishing that mechanical integrity arose from polymer-cell interactions rather than polymer entanglement alone. Confined cells retained modular functionality, as demonstrated by their ability to excrete a recombinant protein and function as biosensors in response to small molecule inducers. Overnight incubation in a fructose solution disassembled macroscopic hydrogels, enabling separate recovery of cells and polymer. Crucially, recovered polymers were spectroscopically indistinguishable from the original material by $^{1}$H NMR and could be redeployed to fabricate new hydrogels. This work demonstrated three design principles applicable beyond the specific chemistry: (i) endogenous cell-surface polyols can serve as natural anchoring points, removing the need for surface modification; (ii) multivalency is a tunable parameter that controls polymer coating density on cell surfaces, aggregate formation, and bulk modulus; and (iii) dynamic bond exchange enables disassembly and recyclability in living materials.

An unexpected phenomenon emerged that pointed to a level of programmability absent in conventional abiotic boronate ester systems: when polymer-cell aggregates were incubated with glucose, polymers initially detached from cells but appeared to reattach after about 3 h, as measured by

fluorescence. We reasoned that this anomalous recovery reflected a dynamic interplay between synthetic polymers and living cells, in which cells consumed glucose and shifted the boronate ester equilibrium back toward polymer-cell association as the competitor concentration decreased. In other words, cellular metabolism, not a synthetic design element, drove a feedback loop that restored network connectivity. This observation reframed polymer-cell assemblies not as static composites, but as chemically coupled systems capable of metabolism-driven behavior.

Inspired by this discovery, we set out to build macroscopic dissipative living materials in which bacterial metabolism itself drives phase transition (Figure 3b).[30] Synthetic dissipative materials have been engineered to consume fuel and produce transient structural responses, but capturing the dynamic livingness of biological materials remains challenging. Biological machinery is complex, metabolic routes are diverse, and the interactions between cells and their molecular environment are difficult to reduce to tractable design principles. We envisioned sugar-fueled dissipative living materials as a tractable, metabolism-driven model system that cycles through sugar-boronic acid displacement for disassembly and cellular metabolic sugar consumption for reassembly.

Central to this design is the choice of living crosslinker. *Staphylococcus epidermidis*, a non-sporulating gram-positive bacterium bearing teichoic acid-rich cell walls, was selected for its tolerance of various osmotic pressures and sustained metabolic activity under the conditions required for polymer-cell assembly (Figure 3b). Macroscopic living hydrogels formed upon mixing aqueous solutions of the cells and APBA polymer at pH 8.8. Incubation in D-glucose (30 °C, 3 h) collapsed the network into a suspension of cell clusters, consistent with disruption of boronate ester crosslinks. Suspensions of disassembled materials reassembled into a gel after an additional 14 h, consistent with a metabolically driven decrease in extracellular D-glucose concentration. However, the storage modulus was reduced by approximately an order of magnitude relative to the original gel. The reassembled gel had also acidified to pH 6.8, significantly lower than the initial pH of 8.8, consistent with the accumulation of metabolic organic acids that shift the boronate ester equilibrium toward the unbound state. After re-adjusting the pH back to 8.8, the dissipative cycle could be repeated, but the stiffness of the second reassembled material remained diminished even after pH restoration. Assays monitoring extracellular D-glucose concentration at different stages of the dissipative cycle revealed a 92% reduction over 17 h during the first cycle and only a 58% reduction during the second cycle, directly implicating diminished metabolic activity in the loss of mechanical recovery.

These results establish a general design principle: when stimuli-responsive bonds are used to couple polymers to metabolically active cells, the cells can act as chemical operators embedded in the network. Metabolic activity continuously perturbs the bond equilibrium, making the state of the material a function of both chemistry and biology. This principle generalizes beyond boronate esters; any dynamic bond whose equilibrium is sensitive to metabolite concentration, pH, or redox state is a candidate for metabolism-driven material behavior. Yet this metabolic coupling comes with inherent constraints. Cells require nutrient-rich, aqueous environments, which limit the accessible materials landscape to hydrated, low-energy applications. Bacterial spores, which survive material processing conditions incompatible with vegetative cells, offer a route to expand this landscape (Figure 1c).

## Bacterial spores expand the scope of ELM design

When bacteria experience nutrient limitation, oxidative stress, or other environmental pressures, some species commit to an asymmetric cell division that produces the metabolically dormant endospore.[31, 32] The mother cell assembles protective layers of macromolecules around the developing spore, conferring tolerance to elevated temperatures, organic solvents, extreme pH, dehydration, and irradiation (Figure 4).[33] Crucially, dormant spores can be germinated on demand to restore full metabolic and cellular function, making spore-forming bacteria uniquely suited as living components that survive material fabrication while retaining biological programmability.

The choice between vegetative cells and spores as living components involves fundamental trade-offs that directly shape polymer design strategy. Vegetative cells maintain continuous metabolic activity: they sense, respond, and generate outputs in real time, making them the preferred chassis for ELMs that require active biosensing, continuous bioproduction, or metabolism-driven dynamic behavior such as dissipative hydrogels. However, this activity is inseparable from the need for a tightly controlled microenvironment, including water availability, physiological pH, nutrient access, and protection from toxic solvents or reactive monomers, which generally restricts polymer processing to mild, aqueous, and low-energy conditions.

Spores, by contrast, are metabolically dormant and can survive harsh processing conditions that are incompatible with vegetative cells. This resilience enables a broader range of processing conditions, storage options, and polymer chemistries than those possible in cell-based ELMs. The trade-off is functional latency: spore-based materials require germination to recover active metabolism, and germination kinetics and efficiency within polymer matrices introduce additional layers of biological and materials complexity. Dormant spores can also support catalytic function without germination via surface-displayed enzymes, as illustrated in the following subsection.[34] Generally, vegetative cells are better suited for dynamic, metabolism-dependent behavior under mild conditions, whereas spores enable robust and processable materials with on-demand activation or ambient-condition stability.

**Spore-enabled material robustness and processing compatibility**

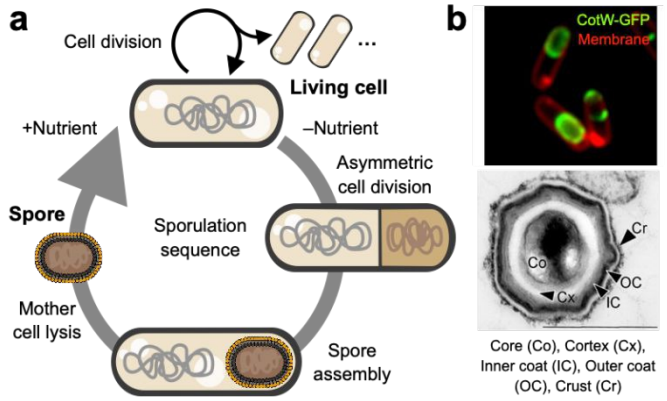


**Figure 4. Lifecycle of Sporulating Cells and Multilayered Structure of Bacterial Spores.** (**a**) General sporulation sequence of bacteria. (**b**) Multilayered spore assembly formed within mother cells. Adapted with permission from ref. 33, Copyright © 2010 Elsevier Ltd.

The durability of spores expands the processing window for material manufacturing. González *et al*. demonstrated this principle by 3D printing agarose with *Bacillus subtilis* spores at 72 °C, a temperature lethal to most vegetative cells.[35] Embedded spores tolerated a broad range of stresses, including high osmolarity, extreme pH, UV light (365 nm), high temperature (80 °C), ethanol, and brief γ-radiation exposure. After a month of dry storage, the spores could be germinated to execute programmed functions such as GFP expression. Leveraging a similar post-fabrication germination strategy, Tang *et al*. reported a self-degrading plastic-spore composite.[36] Germination into vegetative cells triggered lipase secretion to fully degrade poly(caprolactone) (PCL) within one month. This functionality relied on the spores' stress tolerance, allowing them to survive harsh material processing steps such as toluene solvent casting and high-temperature melt extrusion at 120 °C. Importantly, spore incorporation programmed PCL degradation without significantly affecting the plastic films' tensile or thermal properties, consistent with minimal polymer-cell interactions.

The stress tolerance of *B. subtilis* spores can be further enhanced by adaptive laboratory evolution (ALE), a directed approach that cultivates microbes under specific stress conditions for many generations. Kim *et al*. applied ALE to develop a degradable thermoplastic polyurethane (TPU) composite incorporating heat-shock-tolerized spores.[37] These evolved spores withstood hot melt extrusion at 135 °C, retaining 96–100% viability compared to ~20% for wild-type spores. Notably, incorporation of the evolved spores within TPU enhanced toughness by 37%, suggesting that TPU-spore interfacial adhesion improved tensile properties. This mechanical reinforcement implicates polymer-spore interfacial adhesion as a previously underappreciated design variable in spore-based composites.

### Functional spore-based composites and enzyme display

*B. subtilis* spores can be engineered to display surface enzymes,[34] acting as functional filler particles without requiring germination. We developed the TIED platform (Figure 5a), leveraging T7 RNA polymerase to overcome the prior limitations of low enzyme loading density and insufficient catalytic activity in spore surface display.[38-43] TIED utilizes (1) T7 RNA polymerase-driven expression of recombinant proteins at the late stage of sporulation and (2) early harvest of spores from mother cells to avoid nonspecific proteolytic degradation, thereby achieving a high loading density of $10^6$–$10^7$ recombinant enzymes per spore. TIED enzymes are not only recyclable but can be fully renewed after activity loss by inducing spore germination and subsequent re-sporulation (Figure 5b).

We demonstrated the utility of TIED spores displaying a high density of lipases to facilitate enzymatic degradation of polyester in biocomposites.[44] PCL composites were prepared by incorporating engineered spores through dichloromethane solvent casting. As anticipated, the resulting films exhibited self-degrading behavior in aqueous buffer, which was absent in wild-type spore controls. Importantly, the biocomposite retained stiffness and toughness comparable to neat PCL, with only a modest reduction in ductility. Lipase-displaying spores recovered from degraded materials maintained their enzymatic activity and could be reused in subsequent cycles. These results demonstrate that embedding genetically engineered bacterial spores into polymer matrices provides a robust and scalable strategy for installing programmable functionality into synthetic materials without requiring germination into vegetative cells.

### Dynamic covalent assembly with spores enables ELMs with programmable mechanical properties

The chemical durability of *B. subtilis* spores makes them uniquely suited for covalent assembly, enabling systematic engineering of crosslink-dependent stiffness, self-healing, and recyclability. We postulated that spore surface glycans (diols) can participate in dynamic covalent boronate ester crosslinks to afford materials with tunable stiffness (Figure 6a).[45] Spore labeling experiments using fluorescent phenylboronic acid (PBA) probes revealed that binding increased with PBA reactivity, scaling with Hammett substituent constants ($\sigma_{meta}$). Critically, the same trend held in 98% methanol. PBA-spore association is therefore not limited to aqueous conditions, a direct consequence of spore chemical durability that expands the accessible polymer chemistry toolbox beyond what living cells permit.

PBA-functionalized polymers were designed for assembly under both aqueous and non-aqueous conditions. 2-aminoethyl methacrylate (AEMA) provided amine handles for post-polymerization PBA conjugation. 2-hydroxyethyl methacrylate (HEMA) and di(ethyleneglycol) methyl ether methacrylate (DEGMEMA) ensured high solubility. Statistical copolymerization afforded compositions with ~10% PBA content that were highly soluble in methanol. Mixing polymer solutions and spore suspensions afforded water-stable gels, confirming that spores act as multivalent crosslinking nodes to sustain network integrity. Increasing the PBA reactivity increased the gel's storage modulus (Figure 6b), consistent

with greater polymer-spore crosslink density. Notably, the storage modulus increased with both PBA acidity and gel basicity, consistent with stiffening driven by the thermodynamically favored formation of anionic boronate ester crosslinks and establishing PBA p$K_a$ and solution pH as orthogonal handles for modulating gel mechanics.

Reversible crosslinks granted dynamic features. The dynamic equilibrium of boronate ester crosslinks enabled rapid network reorganization, manifesting in self-healing, stimuli-responsive disassembly, and recyclability. The material was cut into two pieces and placed back together in contact, prompting a self-healing response to fully recover the material's viscoelastic properties. In addition, incubation in 50 mM fructose triggered material disassembly through diol competition with spore surface glycans. Solution turbidity increased over 12 h, after which the polymer was recovered quantitatively and redeployed to assemble a new batch of materials.

The dynamic covalent assembly framework positions engineered spores as both structural crosslinking nodes and functional units within the material. The function of the spore itself, defined by recombinant surface enzyme display via TIED, is modular. We therefore envisioned that a hydrogel containing engineered *B. subtilis* spores could act as a catalytic material combining reusability, renewability, and exceptional benchtop stability. Incorporation of APEX2-displaying TIED spores afforded catalytic materials that converted Amplex Red into fluorescent resorufin, and the enzyme's stability within the crosslinked network enabled repeated reuse without loss of activity. The principal limitation of this format is kinetics: mass transport constraints reduce reaction rates relative to free-floating spores or enzymes. However, the spore material offers a distinct operational advantage over live-cell ELMs: it could be stored in abiotic conditions for days without losing catalytic activity. In addition, spores recovered from material disassembly retained full germinability, enabling renewal of functional particles on demand. We note that reaction kinetics slow further after storage, which we attribute to condensation-driven network densification during drying that progressively reduces mesh size and substrate diffusivity. This observation suggests that polymer mesh engineering may become an important design lever for future catalytic ELMs. For example, stimuli-responsive comonomers that transiently swell or expand the network in response to temperature or pH could further enable on-demand transport during catalytic or germination cycles. These concepts illustrate how polymer physics and network architecture may ultimately govern the operational performance of spore-embedding materials.

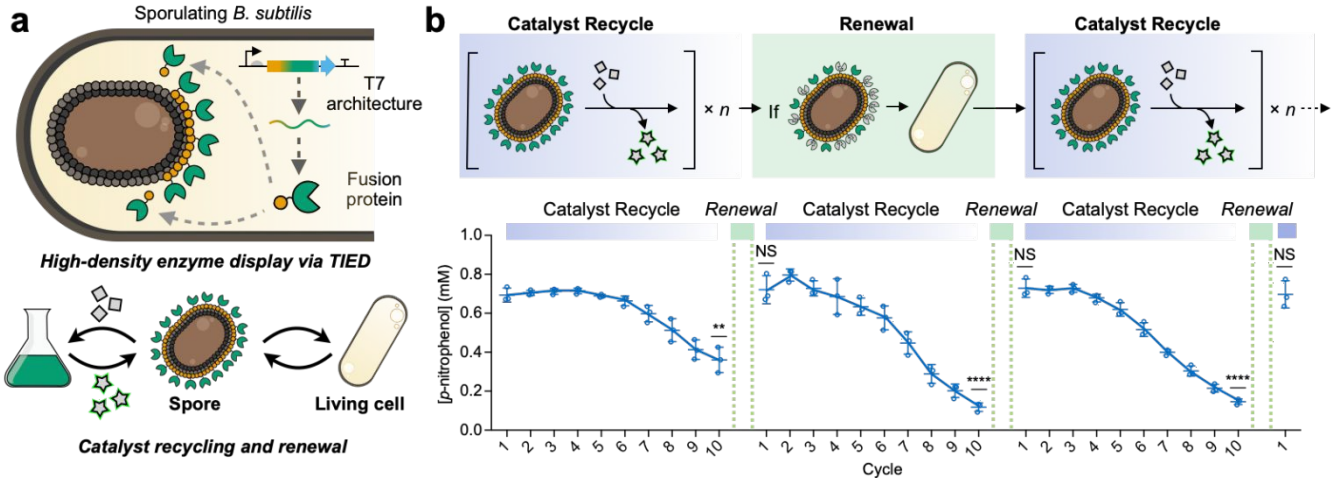


**Figure 5. TIED Platform to Display High Density of Recombinant Enzymes on Bacterial Spores.** (**a**) Schematic illustration of the TIED system leveraging T7 RNA polymerase-driven expression of recombinant fusion proteins. (**b**) Recyclable and renewable feature of TIED biocatalysts. Adapted with permission from ref. 34, Copyright © 2022, American Chemical Society.

*B. subtilis* spores are also well-suited for expanding the material design landscape to dry biocomposites, but chemical incompatibilities with the polymer can lead to significant phase separations and undesirable mechanical properties. Without sufficiently strong and multivalent interactions, the material may exhibit biotic-abiotic phase separations and uncontrolled spore release. We envisioned that dynamic covalent polymer-spore assemblies in organic solvents could mitigate these issues through polymer-spore crosslinking to afford mechanically robust and functional biocomposites. Cysteine residues on *B. subtilis* spores were targeted with benzalcyanoacetate (BCA)-functionalized polymers to form dynamic covalent thia-Michael (tM) bonds in organic solvents (Figure 6c).[46] The formation of BCA-spore bonds was validated through fluorescent labeling, $^1$H NMR, and Raman spectroscopy, all of which revealed tM adducts that were tunable through the electronic contribution of the BCA aryl substituent.

Polymer design was guided by two independent molecular handles. BCA reactivity governed crosslink density and exchange kinetics, while comonomer side chains controlled solvophilicity and backbone rigidity. BCA methacrylate monomers bearing various aryl substituents were copolymerized with methyl methacrylate (MMA) or ethylene glycol methyl ether methacrylate (EGMEMA) to afford polymers

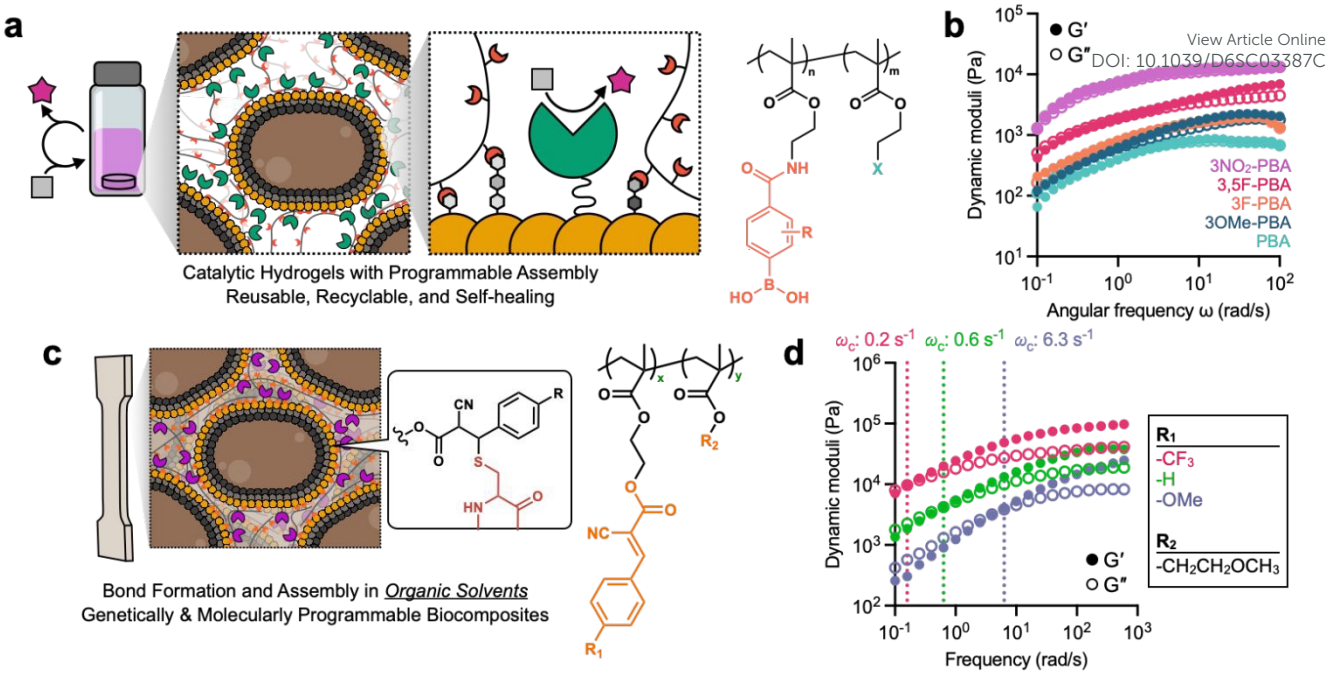


**Figure 6. Dynamic Covalent Assembly with Spores Enables ELMs with Programmable Mechanical Properties.** (**a**) Catalytic hydrogels produced by assembling PBA-functionalized polymers with engineered *B. subtilis* spores displaying recombinant enzymes. Spore surface glycans (bearing diols) form dynamic covalent bond with PBA-functionalized polymers, forming boronate ester crosslinks. Adapted with permission from ref. 45, Copyright © 2023, American Chemical Society. (**b**) Storage and loss moduli of assembled materials at 25 °C. Adapted with permission from ref. 45, Copyright © 2023, American Chemical Society. (**c**) Mechanically robust and functional biocomposites leveraging abundant cysteines on spore surface and thia-Michael bond formation. Adapted from ref. 46, Copyright © 2025 The Authors. (**d**) Viscoelastic behavior of materials. Adapted from ref. 46, Copyright © 2025 The Authors.

of high and low $T_g$, respectively. Mixing DMSO solutions of the polymers with lyophilized spores afforded organogels with programmable stiffness and stress relaxation (Figure 6d). MMA-based materials were consistently stiffer than EGMEMA-based materials. Hansen solubility parameter analysis attributed this to better solubility matching between EGMEMA polymers and DMSO that promoted plasticization. Rheological measurements of the EGMEMA gels confirmed that both storage modulus and relaxation times scaled systematically with BCA electrophilicity, with relaxation times correlating well with Hammett constants ($\sigma_+$), demonstrating that the electronic profile of polymer-spore crosslinks can program viscoelastic behavior.

Dry composites were prepared by solvent casting DMSO gels of spores and low-$T_g$ EGMEMA polymer, and exchanging the solvent with ethanol to afford dog-bone shaped materials primed for mechanical testing. Tensile tests revealed that only the materials with the reactive BCA variant were mechanically reinforced upon spore incorporation, raising the Young's modulus by ~100 MPa. The same reactivity threshold that controlled stiffness also governed biocontainment: while the most reactive BCA network retained spores in aqueous buffer, other materials showed measurable spore leakage that was inversely correlated with BCA reactivity. Optical and scanning electron microscopy further revealed that polymer-spore interfacial homogeneity tracked with BCA reactivity, with the most reactive variant exhibiting near-continuous interfaces and the least reactive variant showing phase separations into spore-rich and polymer-rich domains.

Unlike the catalytic spore hydrogel, installing catalytic function in the dry biocomposite required reconciling two competing constraints: the organic solvent required for polymer processing and the solvent sensitivity of the recombinant enzyme displayed on spores. Screening APEX2-displaying spores in a panel of solvents revealed a sharp dependence of catalytic activity on solvent type. DMSO caused near-complete loss of function and visible spore morphological collapse, while acetone preserved both spore structure and partial enzymatic activity. This outcome guided the selection of acetone as the assembly solvent for catalytic biocomposites. These materials successfully catalyzed the conversion of Amplex Red to resorufin with full efficiency and could be reused in a second catalytic cycle. The platform was extended to mixed aqueous-organic conditions, where significant catalytic activity was retained at up to 12.5% acetone. Crucially, the renewable character of the spore particle provided a route to complete restoration of catalytic function. Spores recovered from material disassembly were germinated, renewed, and confirmed to display full APEX2 activity indistinguishable from pristine engineered spores. This regeneration cycle is uniquely enabled by the dormancy and genetic tractability of spore particles and is a fundamental design advantage of spore-based biocomposites over materials incorporating vegetative cells or purified enzymes.

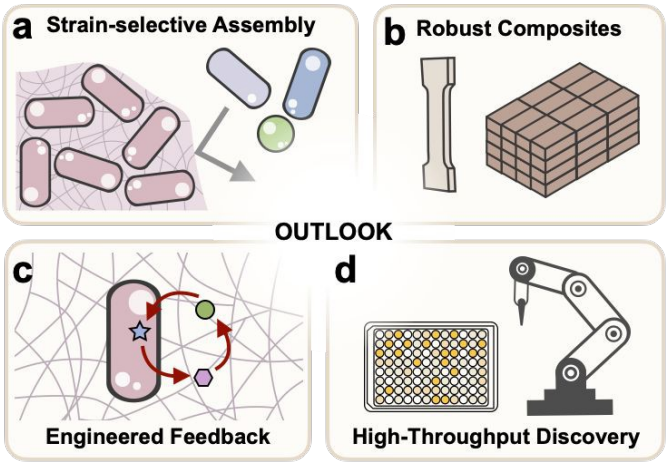


**Figure 7. Opportunities in Polymer-Centered Design at Living-Material Interface.** (**a**) Strain-selective assembly by polymer designs. (**b**) Robust and dry composite materials. (**c**) Synthetic biology-enabled engineered feedback. (**d**) High-throughput discovery platform.

These biohybrid composites establish three generalizable design principles for spore-based ELMs. First, crosslinker reactivity is a molecular handle that governs stiffness, biocontainment, and interfacial homogeneity, a rare convergence of mechanical control and functional design. Second, comonomer identity provides an orthogonal handle for solvophilicity, viscoelasticity, and processibility decoupled from crosslink chemistry. Third, spore renewal through germination and resporulation is a regenerative mechanism unavailable in systems built from purified enzymes and is a fundamental design advantage of genetically tractable biological particles. Together, these principles extend living material design into polymer-processing regimes and operational environments inaccessible to vegetative cells.

## Conclusions and Outlook

The convergence of polymer chemistry and synthetic biology offers opportunities to create new ELM technologies for human health, biomanufacturing, and sustainability. Realizing this potential will require treating the polymer scaffold and living component not as independent design decisions but as an integrated system, one in which molecular interactions at the biotic-abiotic interface actively encode bulk material behavior. The examples highlighted throughout this Perspective illustrate this principle across a range of polymer chemistries and cell types: covalent crosslinks program mechanical robustness and biocontainment; dynamic covalent bonds introduce stimuli-responsiveness and recyclability; and metabolic activity serves as a thermodynamic engine for far-from-equilibrium material behavior. Expanding this design space to bacterial spores further demonstrates that material-compatible biological particles, when molecularly coupled to synthetic polymers through well-defined interfaces, can extend programmable material design across the mechanical and functional range separating synthetic networks from living materials.

A central message of this Perspective is that polymer chemistry is an essential design toolkit for living materials, not merely a passive scaffold for cells. Molecular interactions at the polymer-cell interface encode mechanical properties, transport, stimuli-responsiveness, and dynamic behavior. Realizing this potential more broadly will require confronting the current bottlenecks that constrain the field, such as nutrient dependence and pH drift during metabolic coupling cycles, mass-transport limitations that slow reaction kinetics in dense or dry networks, and durability loss across repeated functional cycles as cellular viability diminishes. Each of these represents a polymer design challenge – network engineering to control transport, buffer-capacity-matched polymer chemistries to resist pH excursions, and stimulus-triggered nutrient release to extend operational lifetime – pointing toward materials designs for the next phase of ELM development.

Advances in polymer science will expand the design space of next-generation ELMs, enabling more sophisticated material architectures without compromising the functionality of the living component. A deeper understanding of how polymer composition, architecture, and network dynamics influence cellular and macroscopic material behavior will enable the design of integrated systems whose performance exceeds the simple sum of their individual components. Biocompatible methods to facilitate synthetic and supramolecular transformations on the cell surface will broaden the chemical vocabulary available for polymer-cell interfacial engineering. Greater material complexity can be achieved through molecularly programmed cell strain specificity. Glycan profile differences and macromolecular diffusion barriers at the cell surface represent underexplored molecular handles for encoding this strain-specific assembly (Figure 7a).

Expanding the chemical vocabulary at the living-material interface will require the development of new polymer platforms. Sequence-defined polymers offer the prospect of information-rich recognition at the polymer-cell boundary. This approach can offer control over cell organization and behavior without requiring cell modifications. Dynamic covalent networks and vitrimers are particularly well-positioned to extend ELM fabrication toward conventional polymer processing. Their combination of structural robustness and thermally or chemically triggered bond exchange could enable melt-processable or moldable living composites, bridging the gap between the solvent-cast spore-based materials and scalable plastics manufacturing (Figure 7b).

Advanced synthetic biology tools to engineer cells and spores to sense, adapt, and contribute to material performance in real time will create new opportunities for materials design. Expanding the scope and chemical precision of engineered genetic circuits that create chemical connections between the inside and outside of living cells could unlock a wide range of dynamic behavior in materials. Specifically, genetic circuits that sense and respond to local polymer and chemical cues could create the feedback loop between synthetic scaffolds and the engineered living component (Figure 7c). Chassis engineering with extremophiles and fungi, which offer distinct surface

chemistries and metabolic repertoires, further extends ELM functionalities and operational environments.

Development of these expanded tools can be accelerated using a high-throughput experimentation platform. Automated testing for materials by varying cells with different genetic circuits, polymer architecture, and processing conditions will systematically map a large and otherwise intractable parameter space. Feeding high-quality datasets derived from these platforms into a design-build-test-learn workflow will ultimately enable predictive engineering of specific ELM functionalities (Figure 7d).

Translating ELMs beyond proof-of-concept demonstrations will require confronting technological barriers that are distinct from those in conventional materials development. Controlling biocontainment will be a prerequisite for any application involving open or semi-open deployment, and polymer chemistry offers concrete design strategies. Covalent encapsulation can prevent cell escape under ambient conditions, while stimuli-triggered network dissolution enables controlled release only under defined conditions. Scalable manufacturing presents a second barrier. The kinetics of polymer-cell assembly, crosslink density, and cell viability under industrial mixing and processing conditions will all need to be characterized as a function of scale. Long-term operational stability – encompassing both polymer degradation, loss of cellular viability, and genetic drift across repeated use cycles – is a third bottleneck that polymer design can partially address through degradation-resistant network architectures, redox- or pH-buffering polymer matrices, and stimulus-triggered nutrient depots that extend operational lifetime. Addressing all three will require closer integration of polymer science and bioengineering expertise, and will likely define the field's applied research agenda over the next decade.

Ultimately, the future of ELMs will depend not only on what living systems can do, but also on how precisely polymer chemistry can direct, stabilize, and amplify those functions at the living-material interface.

## Conflicts of interest

There are no conflicts to declare.

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## Data Availability Statement

No primary research results, software or code have been included and no new data were generated or analysed as part of this review.